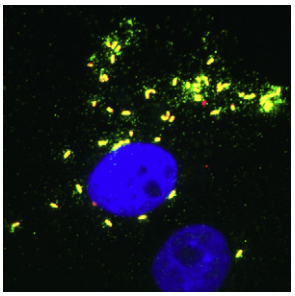
# Microbiology Select

Cell

Microbial pathogens have evolved strategies to live and multiply within their hosts, who in turn have evolved surveillance systems that detect such pathogens and trigger countermeasures. This Microbiology Select highlights recent papers that reveal new aspects of these host and microbial strategies.

## *Salmonella* Intoxicates from within



Photomicrograph of cultured cells infected with *Salmonella* Typhi and stained for typhoid toxin (green), *S.* Typhi (red), and DNA (blue). Image courtesy of S. Spanò.

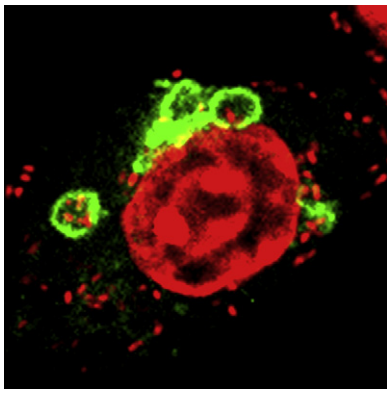
The bacterial pathogen that causes typhoid fever in humans, *Salmonella enterica* serovar Typhi (*S.* Typhi), can colonize its human hosts for their entire lifetime. *S.* Typhi secretes a toxin whose active component is a protein with DNase activity, CdtB. Intoxicated host cells exhibit cell-cycle arrest resulting from DNA damage. Spanò et al. (2008) now show that *S.* Typhi CdtB is part of a multifunctional toxin with a complex evolutionary ancestry, which the authors have named "typhoid toxin." In addition to CdtB, typhoid toxin is composed of subunits PltA and PltB, which are homologs of the ADP ribosylating A subunit of pertussis toxin and one of the components of its B subunit, respectively. The *S.* Typhi multipartite toxin is produced only when the bacterium is within a host cell, but Spanò et al. show that this toxin does not directly intoxicate this host cell. Instead, like a hormone, the toxin acts after it has been secreted and then taken up, either by the infected toxin-producing cell itself or by distant uninfected cells. Indeed, addition of a toxin-neutralizing antibody eliminated the toxicity in infected and uninfected cells. The authors hypothesize that infected cells lacking a receptor for the exported toxin could act as toxin delivery factories. The next step will be to identify the role of typhoid toxin during persistent infection, which may confirm that this unusual mode of exotoxin delivery is key for *S.* Typhi's adaptation to life within a human host.

*S. Spanò et al. (2008). Cell Host Microbe* ***3****, 30–38.*

## *Listeria* Preserves a Refuge

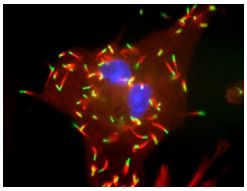
*Listeria monocytogenes* is a bacterial pathogen that causes opportunistic infections in humans and, like other pathogens, secretes a toxin as part of its intracellular lifestyle. *L. monocytogenes* secretes listeriolysin O (LLO), a cholesterol-dependent pore-forming toxin. After entering phagocytic cells, *L. monocytogenes* occupies a vacuole, whereupon LLO perforates the vacuolar membrane, blocking maturation of the vacuole into an antibacterial environment and facilitating escape of bacteria to the hospitable cytosol. Cytosolic bacteria rapidly multiply, ultimately infecting neighboring cells. Now, Birmingham et al. (2008) reveal yet another way in which LLO subverts host cell defenses. They show that a subpopulation of intracellular *L. monocytogenes* does not escape to the cytosol but can be found slowly replicating within large non-degradative vacuoles. They provide evidence that this restriction is due to host factors partially inhibiting LLO activity. The reduced LLO activity is insufficient for bacteria to escape to the cytosol but sufficient to preserve the vacuole by counteracting the host autophagy pathway for recycling damaged cellular components. Although reducing LLO toxin activity contains the *L. monocytogenes* infection, these bacteria persist within the host. Such a balance between bacterial toxin activity and host defenses, shifting over evolutionary time, may help to explain the diversity of infectious disease outcomes.

*C. Birmingham et al. (2008). Nature* ***451****, 350–354.*



*Listeria monocytogenes* (red dots) inhabits spacious vacuolar compartments (green) in macrophages after infection (DNA, red). Image courtesy of C. Birmingham.

## Host Cells Pinpoint the Enemy

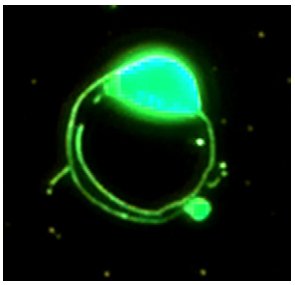


*Listeria monocytogenes* (green) polymerizing host actin (red) in the cytosol of an infected macrophage. Image courtesy of C. Rae.

Macrophage infection experiments with wild-type *Listeria monocytogenes* bacteria and with mutants lacking the pore-forming listeriolysin O toxin, if appropriately timed, yield two predominant bacterial populations, one that has escaped to the host cell cytosol and one that remains trapped inside vacuoles. Leber et al. (2008) now use these separate populations of *L. monocytogenes* to examine how phagocytic cells respond to infection based on location of the bacterial populations within the cell. Using DNA microarray analysis, the authors identified seven host cell genes whose regulation is directly altered specifically by the presence of cytosolic (as opposed to vacuolar) *L. monocytogenes*. The altered expression of one of these genes, which encodes the defensive cytokine interferon β, could be recapitulated by introducing *L. monocytogenes* DNA into the cytosol. Indeed, there was synergy between the response to DNA and the response to a cell wall component of the bacterium codelivered to the cytosol, suggesting that this host signaling pathway integrates responses to individual molecular signatures. By sensing both the pathogen composition

and location, this host cell pathway could be used to trigger defenses tailored to the pathogen and to the stage of its intracellular life cycle. Analysis of the host response specific to the subpopulation of *L. monocytogenes* persisting within nondegradative vacuoles might yield clues about the nature of chronic bacterial infections.
*J.H. Leber et al. (2008). PLoS Pathog.* ***4****, e6. 10.1371/journal.ppat.0040006.*

## *Toxoplasma* Tips Off Its Host

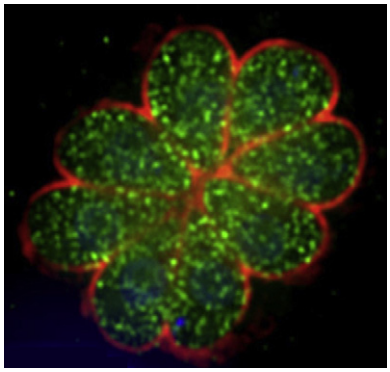


**Indirect immunofluorescence showing the *Toxoplasma gondii* surface antigen (green) left behind as the parasite glides in circles on a glass slide. Image courtesy of F. Plattner.**

*Toxoplasma gondii* is a protozoan parasite that opportunistically infects humans. Like the bacteria *Salmonella enterica* serovar Typhi and *Listeria monocytogenes*, this parasite is an intracellular pathogen and must be able to withstand defenses triggered by host surveillance systems. *T. gondii* uses actin-based motility to invade host cells, and now Plattner et al. (2008) show that *T. gondii* profilin, a protein that regulates actin polymerization, is required for motility of this parasite and for its ability to invade and actively exit host cells. *T. gondii* profilin is recognized by Toll-like receptor TLR11, a component of the host innate immune system. In this respect, parasite profilin resembles bacterial flagellin, another microbial motility protein that is recognized by the host innate immune system. Plattner et al. demonstrate that profilin is required for TLR11-dependent production of the defensive host cytokine interleukin-12. *T. gondii* cells in which the endogenous profilin was replaced by malaria parasite profilin retained their motility and their ability to invade host cells; however, they no longer triggered the defensive interleukin-12 host response. Why then has *T. gondii* not evolved a profilin that can evade detection by the host immune system? There may be selection pressures that constrain the evolution of *T. gondii* profilin and that have implications for understanding the ongoing evolutionary arms' race between parasites and their hosts.
*F. Plattner et al. (2008). Cell Host Microbe* ***3****, 77–87.*

## *Toxoplasma* Telegraphs Its Exit

The protozoan pathogen *Toxoplasma gondii* and plants share a calcium-mediated signaling pathway regulated by the second messenger cyclic ADP ribose. In plants, cyclic ADP ribose is regulated by the hormone abscisic acid. Now, Nagamune et al. (2008) show that abscisic acid serves this function in *T. gondii* as well. *T. gondii* produces abscisic acid most likely by using biosynthetic pathways operating in the apicoplast, a parasite organelle derived from an ancient algal endosymbiont. Abscisic acid levels peak in *T. gondii* after it has replicated inside the host cell and just before it emerges to invade new host cells. The herbicide fluridone is known to inhibit the abscisic acid biosynthetic pathway in plants. It also does so in *T. gondii*, blocking the parasite's escape from its host cell and reducing *T. gondii* dissemination in mice and so preventing lethal infection. Thus, abscisic acid controls calcium signaling in *T. gondii* and egress of the parasite from infected host cells. *T. gondii* production of a plant hormone provides an opportunity to use knowledge of plant signaling pathways to identify new targets for drugs for combating *T. gondii* infections and perhaps also those caused by the related protozoan, the malaria parasite.
*K. Nagamune et al. (2008). Nature* ***451****, 207–210.*



**An intracellular cluster of *Toxoplasma gondii* is outlined by a surface marker (red) and filled with a cytoplasmic marker (green). Image courtesy of J. Gordon.**

**David A. D'Argenio**